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Atty. Ref.: 117-319
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Supplemental Amendment

IN THE CLAIMS:

Amend the claims as follows:

Claims 1-49. (Canceled)

50. (New) An expression vector which comprises:

- (a) a polynucleotide comprising the entire nucleotide sequence set out in SEQ ID NO. 1 or the complement thereof;
- (b) a polynucleotide comprising a polynucleotide sequence which is degenerate as a result of the genetic code to the polynucleotide of SEQ ID NO. 1; or
- (c) a polynucleotide that encodes a polypeptide that comprises:
 - (i) the sequence set out in SEQ ID NO: 2;
 - (ii) the polypeptide encoded by nucleotides 210-1335 of SEQ ID NO: 1;
 - (iii) amino acids 158-211 of SEQ ID NO: 2; or
 - (iv) amino acids 380-444 of SEQ ID NO: 2,

wherein said polypeptide has the ability to stimulate an immune response against the polypeptide of SEQ ID NO: 2;

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operably linked to regulatory sequences capable of directing expression of said polynucleotide in a host cell.

51. (New) A vector according to claim 50 which is a plasmid or viral vector and wherein said polynucleotide is under control of a promoter.

52. (New) A plasmid according to claim 51, wherein the promoter is a CMV, MMLV, RSV or SV40 promoter.

53. (New) A pharmaceutical composition comprising:

- (a) a polynucleotide selected from:
 - (i) a polynucleotide comprising the entire nucleotide sequence set out in SEQ ID NO: 1 or the complement thereof;
 - (ii) a polynucleotide comprising a polynucleotide sequence which is degenerate as a result of the genetic code to the polynucleotide of SEQ ID NO: 1; and
 - (iii) a polynucleotide which encodes a polypeptide as defined in (a); or
- (b) a polypeptide which comprises:
 - (i) the sequence set out in SEQ ID NO: 2;
 - (ii) the polypeptide encoded by nucleotides 210-1335 of SEQ ID NO: 1;

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(iii) amino acids 158-211 of SEQ ID NO: 2; or

(iv) amino acids 380-444 of SEQ ID NO: 2;

wherein said polypeptide has the ability to stimulate an immune response against the polypeptide of SEQ ID NO: 2,

and a suitable carrier or diluent.

54. (New) A method of treating or preventing infection by *Mycobacterium avium* subspecies *paratuberculosis* (MAP) or a disease caused by MAP in an animal or human which method comprises administering to the animal or human an effective amount of:

(a) a polynucleotide selected from:

(i) a polynucleotide comprising the entire nucleotide sequence set out in SEQ ID NO: 1 or the complement thereof;

(ii) a polynucleotide comprising a polynucleotide sequence which is degenerate as a result of the genetic code to the polynucleotide of SEQ ID NO: 1; and

(iii) a polynucleotide which encodes a polypeptide as defined in (a); or

(b) a polypeptide which comprises:

(i) the sequence set out in SEQ ID NO: 2;

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(ii) the polypeptide encoded by nucleotides 210-1335 of SEQ ID NO: 1;

(iii) amino acids 158-211 of SEQ ID NO: 2; or

(iv) amino acids 380-444 of SEQ ID NO: 2;

wherein said polypeptide has the ability to stimulate an immune response against

the polypeptide of SEQ ID NO: 2.

55. (New) A method according to claim 54, wherein the disease is Johne's disease or Crohn's disease.

56. (New) A vaccine composition comprising

(a) a polynucleotide selected from:

(i) a polynucleotide comprising the entire nucleotide sequence set out in SEQ ID NO: 1 or the complement thereof;

(ii) a polynucleotide comprising a polynucleotide sequence which is degenerate as a result of the genetic code to the polynucleotide of SEQ ID NO: 1; and

(iii) a polynucleotide which encodes a polypeptide as defined in (a); or

(b) a vector according to claim 50; or

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(c) a polypeptide which comprises:

- (i) the sequence set out in SEQ ID NO: 2;
- (ii) the polypeptide encoded by nucleotides 210-1335 of SEQ ID NO: 1;
- (iii) amino acids 158-211 of SEQ ID NO: 2; or
- (iv) amino acids 380-444 of SEQ ID NO: 2;

wherein said polypeptide has the ability to stimulate an immune response against the polypeptide of SEQ ID NO: 2,

together with a pharmaceutically acceptable carrier or diluent.

57. (New) A nucleic acid vaccine comprising (i) a plasmid as defined in claim 51 and (ii) a pharmaceutically acceptable carrier or diluent.

58. (New) A vaccine according to claim 57 which further comprises a transfection agent.

59. (New) A vaccine comprising:

(a) a polypeptide comprising:

- (i) the sequence set out in SEQ ID NO: 2;
- (iii) the polypeptide encoded by nucleotides 210-1335 of SEQ ID NO: 1;

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(iii) amino acids 158-211 of SEQ ID NO: 2; or

(iv) amino acids 380-444 of SEQ ID NO: 2;

wherein said polypeptide has the ability to stimulate an immune response against the polypeptide of SEQ ID NO: 2;

optionally linked to a hapten molecule, and

(b) a pharmaceutically acceptable carrier or diluent.

60. (New) An isolated non-pathogenic microorganism or a cell isolated from a human or animal species prone to infection by *Mycobacterium avium* subspecies *paratuberculosis* (MAP) which has been transformed or transfected with a nucleic acid construct comprising a polynucleotide selected from:

(a) a polynucleotide comprising the entire nucleotide sequence set out in SEQ ID NO: 1 or the complement thereof;

(b) a polynucleotide comprising a polynucleotide sequence which is degenerate as a result of the genetic code to the polynucleotide of SEQ ID NO: 1; or

(c) a polynucleotide which encodes a polypeptide comprising:

(i) the sequence set out in SEQ ID NO: 2;

(ii) the polypeptide encoded by nucleotides 210-1335 of SEQ ID NO: 1;

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(iii) amino acids 158-211 of SEQ ID NO: 2; or

(iv) amino acids 380-444 of SEQ ID NO: 2;

wherein said polypeptide has the ability to stimulate an immune response against the polypeptide of SEQ ID NO: 2,

or a vector as defined in claim 51.

61. (New) An isolated non-pathogenic microorganism according to claim 60 which is a recombinant bacterium or virus.

62. (New) An isolated non-pathogenic microorganism or a cell according to claim 60, wherein the nucleic acid construct further comprises a polynucleotide which encodes the polypeptides of the GS region of MAP.

63. (New) An isolated non-pathogenic microorganism or a cell according to claim 60, wherein the gene or genes present in the nucleic acid construct are expressed.

64. (New) A vaccine comprising (i) a non-pathogenic microorganism or a cell according to claim 60 and (ii) a pharmaceutically acceptable carrier or diluent.

65. (New) A method of detecting the presence or absence of immunity against *Mycobacterium avium* subspecies *paratuberculosis* (MAP) in an animal or human, which comprises:

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- (a) providing a polypeptide comprising:
 - (i) the sequence set out in SEQ ID NO: 2;
 - (ii) the polypeptide encoded by nucleotides 210-1335 of SEQ ID NO: 1;
 - (iii) amino acids 158-211 of SEQ ID NO: 2; or
 - (iv) amino acids 380-444 of SEQ ID NO: 2;

wherein said polypeptide has the ability to stimulate an immune response against the polypeptide of SEQ ID NO: 2;

- (b) incubating a sample from said animal or human with said polypeptide under conditions which allow for an immune reaction; and
- (c) determining whether any immune reaction against said polypeptide has occurred.

66. (New) A method according to claim 65 for detecting the presence or absence of an antibody capable of binding MAP in a biological sample which method comprises:

- (a) incubating a biological sample with said polypeptide under conditions which allow for the formation of an antibody-antigen complex; and

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(b) determining whether an antibody-antigen complex comprising said polypeptide is formed.

67. (New) A method according to claim 65 for detecting the presence or absence of cell mediated immune reactivity to MAP in an animal or human, which method comprises:

(a) incubating a cell sample with said polypeptide under conditions which allow for cellular immune response; and

(b) detecting the presence of said cellular immune response in the incubate.

68. (New) A method for measuring the response of an animal or human to vaccination against *Mycobacterium avium* subspecies *paratuberculosis* (MAP) comprising detecting the presence or absence of immunity against MAP according to the method of claim 65 in biological samples obtained from said human or animal during and/or after said vaccination.

69. (New) A method according to claim 68, wherein said vaccination is with the vaccine of claim 56.

70. (New) A method according to claim 65 or 68, wherein said sample is selected from blood, milk and saliva.

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71. (New) A method according to claim 65 or 68, wherein said polypeptide carries a revealing label.

72. (New) A method according to claim 65 or 68, wherein said polypeptide is immobilised on a solid support.

73. (New) A polypeptide in isolated form which comprises the sequence set out in SEQ ID No. 2, wherein said polypeptide has the ability to stimulate an immune response against the polypeptide of SEQ ID NO: 2.

74. (New) An isolated polynucleotide selected from:

- (a) a polynucleotide comprising the entire nucleotide sequence set out in SEQ ID No. 1 or the complement thereof;

- (b) a polynucleotide comprising a polynucleotide sequence which is degenerate as a result of the genetic code to the polynucleotide of SEQ ID No. 1; or

- (c) a polynucleotide which encodes the polypeptide of SEQ ID NO: 2, wherein said polynucleotide encodes a polypeptide that has the ability to stimulate an immune response against the polypeptide of SEQ ID NO: 2.